Efficacy of Four Trap Types for Detecting and Monitoring *Culex* spp. in North Central Florida

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ABSTRACT The effectiveness of four types of mosquito traps at sampling *Culex* mosquitoes was compared at two north central Florida study sites (a commercial dairy and a swine research unit) with highly eutrophic lagoons that have a history of producing large populations of *Culex* mosquitoes. Traps evaluated included a John Hock (JH) model 1012 CDC style light trap, JH model 1712 CDC Gravid Trap (GT), and American Biophysics Corporation's Mosquito Magnet-Experimental (MM-X) and commercial model MM-Professional (MM-Pro) traps. The MM-X and MM-Pro traps are based on new counterflow technology. *Culex nigripalpus* Theobald and *Culex quinquefasciatus* Say were the two most abundant species collected, and they were the dominant species at the dairy site and swine research unit, respectively. *Culex erraticus* Dyar & Knabb and *Culex salinarius* Coquillett also were caught but in much lower numbers than the two dominant species at the swine unit, and rarely at the dairy site. At the dairy site the MM-Pro collection of total female *Cx. quinquefasciatus* was significantly greater than all other trap types (MM-Pro > GT > MM-X = CDC), and the MM-Pro also caught the most *Cx. nigripalpus*, but not significantly more than the CDC or MM-X (MM-Pro = CDC = MM-X > GT). At the swine research unit the largest number of female *Cx. quinquefasciatus* were caught in the MM-X trap, but the only significant difference was with the CDC trap (MM-X = MM-Pro = GT > CDC). For *Cx. nigripalpus* the CDC trap caught the most females, but these collections were only significantly different from the MM-Pro and GT (CDC = MM-X > MM-Pro > GT). The GT caught significantly more gravid female *Cx. quinquefasciatus* than all the other trap types at both sites, but collected very few gravid *Cx. nigripalpus* at either site.

KEY WORDS *Culex*, surveillance, West Nile virus, traps, counterflow geometry

West Nile virus (family *Flaviviridae*, genus *Flavivirus*, WNV) is a mosquito-borne zoonosis infecting humans, birds, horses, and other mammals, such as dogs, bats, rodents, rabbits, cats, raccoons, and skunks. It first occurred in the United States in 1999, in New York City, but it has since rapidly spread across the country. A flavivirus native to Africa and the Mideast, it is related to Japanese encephalitis in Asia and St. Louis encephalitis (SLE) in North America. It infects an unusually wide variety of mosquito species, and asymptomatic birds may serve as reservoirs. Mosquitoes in the genus *Culex* are implicated as primary vectors of WNV and SLE virus (family *Flaviviridae*, genus *Flavivirus*, SLEV) (Day 2001, Sardelis et al. 2001, Turell et al. 2001, Goddard et al. 2002). The principal vectors of SLE virus in the eastern United States (except Florida) are thought to be *Culex restuans* Theobald and *Culex pipiens* L. (Mitchell et al. 1980). Both species have now been implicated in WNV transmission and are common in urban areas and some rural ecotopes. Evidence is emerging that *Culex salinarius* Coquillett also may be an important species in the WN virus transmission cycle in North America (Andreadis et al. 2004). In Florida, *Culex nigripalpus* Theobald is the principal vector of SLEV. It is suspected that this species and *Cx. quinquefasciatus* Say will be the principal vectors of WNV in Florida. *Cx. salinarius,* because of its broader feeding habits, may play an important role as a bridge vector to horses and humans.

Until the advent of WNV in the United States, relatively few resources were spent on developing new surveillance or control techniques for *Culex* spp. compared with that spent on major nuisance species such as *Ochlerotatus taeniorhynchus* (Wiedemann). Therefore, the primary objective of this study was to evaluate the relative efficacy of four types of traps for collection of natural populations of *Culex* spp. in north central Florida. Another objective was to compare the relative efficacy of these different adult trapping techniques to capture these *Culex* mosquitoes in various physiological states.

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Materials and Methods

Study Sites. Trapping studies were conducted at two sites, a swine research facility at the University of

Florida, Alachua Co. (29° 37′ N, 29° 37′ W) and a commercial dairy farm in Marion Co. (28° 58′ N, 82° 24′ W). At each site, there was a highly eutrophic lagoon, containing livestock effluent, surrounded by a variety of vegetation types. The vegetation surrounding the lagoon at the swine research facility consisted primarily of oak (*Quercus* spp.); elderberry, *Sambucus canadensis* L.; wax myrtle, *Myrica cerifera* (L.); *Solanum* spp.; and palmetto palm, *Sabal palmetto* (Walt.) Lodd. ex JA & JH Schultes. The lagoon at the commercial dairy farm was surrounded by grass (*Digitana* spp.), sedges (*Cyperus* spp.) and herbaceous vegetation (primarily *Ambrosia artemisiifolia* L., *Amaranthus hybridus* L., and *Ipomoea trichocarpa* Dennst.). At both sites, vegetation was cleared adjacent to traps and traps placed 2–3 m from the edge of the lagoon.

Trap Types. Four trap types were used in these studies. Two trap types have been used for years (Centers for Disease Control [CDC]-type miniature light trap and CDC gravid traps) and two that use relatively new counterflow technology (Kline 1999, Kline 2002). The CDC type trap used was the New Standard Miniature Light Trap-model 1012 (John Hock Company, Gainesville, FL), which uses an incandescent light (CM47 bulb) source. In addition to light the traps were baited with CO_2 (500 ml/min). In these studies, the CDC traps were suspended from a pole so that the top opening was ≈1.5 m above ground level. The CDC Gravid Trap-model 1712 (GT) (John Hock Company) also was used at both study sites. In the GT design, based on a trap designed by Reiter (1983) to collect gravid *Culex*, mosquitoes were drawn from the water surface into a net bag with a 6-V fan in a PVC tube, which was suspended over a pan containing hay infusion as an oviposition attractant. Each GT used a small green pan (22 cm in width by 34 cm in length by 17 cm in depth, Rubbermaid 439, Rubbermaid Commercial Products, Winchester, VA) to contain the hay infusion. These pans were preconditioned by filling them with water and letting them sit for at least a week. They were then scrubbed, rinsed, and dried before use. The infusion was prepared by adding ≈0.5 kg (≈1.1 lb) of hay per 113.56 liters (30 gal) of water. The infusion was incubated for 7 d before use and then used for no more than seven consecutive days. A new batch was started every 7 d. Two liters of the infusion were added to the GT. Each day, the previous day's infusion was discarded, and a fresh 2 liters was added. Both the model 1012 and model 1712 traps operated with a single 6-V, 10-A-h rechargeable gel cell battery, which was replaced daily.

The Mosquito Magnet-Experimental (MM-X, aka CFG or pickle jar trap, American Biophysics Corporation, North Kingston, RI) uses a counterflow concept to attract and capture mosquitoes (Kline 1999). The trap is powered by two 6-V, 10-A batteries connected in series. The MM-X trap was hung from a pole so that the bottom of the attractant plume discharge tube was ≈50 cm (19.69 in.) above ground level. In these studies, the only attractant used by the MM-X trap was CO_2 (500 ml/min).

The fourth trap type used was the commercial model of the Mosquito Magnet-Professional (MM-Pro) (American Biophysics Corp., North Kingston, RI). As with the MM-X trap, the MM-Pro uses a similar counterflow technology to capture insects. Propane gas supplied by 20-lb tanks powered (through catalytic combustion a thermoelectric generator used excess heat to produce electricity) the fan motors, produced heat, moisture, and generated CO_2 (a byproduct of combustion). The trap hung from a wheeled stand that placed the opening ≈60 cm above the ground.

Carbon dioxide for both the CDC model 1012 and MM-X traps was supplied from 9-kg (20-lb) compressed gas cylinders. A flow rate of 500 ml/min was used for both trap types. Control of CO_2 flow rate was achieved with a FLOWSET1 pressure regulator (Clarke Mosquito Control, Roselle, IL) with an output fixed at 15 psig, a 10-μl line filter, a 500 ml/min flow control orifice, and quick-connect luer fittings. Because previous studies showed that octenol might be a repellent to some *Culex* species (Kline 1994), this attractant was not used in these trials.

The traps were operated for ≈24 h. It took ≈15 min each day to rotate them to the next position. Collection nets from all the traps were replaced daily. Collection nets were returned to the laboratory and placed into a regular chest freezer for at least 4 h before their contents were sorted to species and sex. Females were classified as to gonotrophic condition and counted. Female gonotrophic condition was classified following a slight modification of Edman et al. (1975) as blooded if they contained blood but no developed eggs; gravid if they contained fully developed eggs and no evidence of blood; and blood-gravid with partially digested blood and some developed eggs, no matter what the proportion was between blood and developed eggs.

Experimental Design. The experimental design used at both the swine unit and dairy farm was a 4 by 4 Latin square (Steel and Torrie 1980). Within each Latin square, there were four rows (days) each comprising one of four consecutive 24-h periods, and within each row, four blocks (positions), each comprising a separate physical location for each of four types of mosquito traps (treatments). The four traps, one of each type, were placed around the perimeter of each lagoon so that no trap was located closer than 33 m of another trap. Each day, they were rotated clockwise to the next position. Four replicates were completed at Hidden Hammock dairy between 18 June and 7 August 2001; seven replicates were completed at the swine research unit between 18 June and 28 September 2001. Replicates from Hidden Hammock and the swine research unit were analyzed separately.

Statistical Analysis. Each site was analyzed separately. Replicate Latin squares were treated as a split plot on the original Latin square design for whole plots, with days as whole plots and trap positions as subplots. The mosquitoes collected by each trap on each day were counted and categorized, by replicate,

Table 1. Comparisons of four different trap types for the collection of *Culex* mosquitoes at Hidden Hammock Dairy, Marion Co., FL

Species	Sex/physiological state	Mean no. (SE) of mosquitoes collected/trap night for each trap type			
		GT	CDC	MM-X	MM-Pro
Cx. quinquefasciatus	♂/total	24.44 (5.92)a	10.13 (3.18)b	29.31 (8.31)a	87.63 (26.30)c
	♀/total	196.0 (31.72)a	95.25 (24.51)b	104.25 (22.98)b	701.44 (224.41)c
	♀/blooded	2.44 (0.63)a	5.69 (4.42)a	4.13 (1.46)a	30.13 (9.42)b
	♀/blood/gravid	5.38 (3.64)a	0.44 (0.38)b	0.25 (0.14)b	0.69 (0.34)ab
	♀/gravid	73.75 (19.15)a	8.0 (5.52)b	3.69 (1.09)b	8.69 (3.21)b
Cx. nigripaplpus	♂/total	0.50 (0.50)a	0.13 (0.13)a	0.00 (0.00)a	0.06 (0.06)a
	♀/total	1.56 (0.71)a	30.56 (12.19)b	20.13 (6.98)b	59.31 (37.14)b
	♀/blooded	0.00 (0.00)a	0.06 (0.06)a	0.25 (0.19)a	0.19 (0.14)a
	♀/blood/gravid	0.00 (0.00)a	0.00 (0.00)a	0.00 (0.00)a	0.00 (0.00)a
	♀/gravid	0.44 (0.32)a	1.75 (1.56)a	0.81 (0.33)a	0.38 (0.26)a

PROC GLM and multiple comparison (Tukey's HSD) test (SAS Institute 1988) performed after $\log_{10}(x + 1)$ transformations. Means within each row (for each sex and physiological state) having the same letter are not significantly different ($n = 16$ nights, $P \leq 0.05$).

according to species and sex. Females were categorized as unfed (empty), blooded, blooded/gravid, and gravid. Count data were analyzed separately by species within each location using analysis of variance procedures (PROC GLM) (SAS Institute 1988). To minimize heteroscedasticity we transformed each count datum to $\log10(x + 1)$ before analysis (Steel and Torrie 1980). Means separation was made using Tukey's honestly significant difference (HSD) test ($P = 0.05$) (SAS Institute 1988).

Results

Cx. nigripalpus, *Cx. quinquefasciatus*, *Culex erraticus* Dyar & Knabb, and *Cx. salinarius* were captured at both the Hidden Hammock dairy and the University of Florida Swine Research Unit (swine unit) study sites.

Hidden Hammock Dairy. *Cx. quinquefasciatus* was the most frequently trapped species (Table 1). On average, MM-PRO captured significantly more male ($F_{3, 27} = 10.28$; $P = 0.0088$) and female ($F_{3, 27} = 7.25$; $P = 0.0202$) *Cx. quinquefasciatus* than other trap types, whereas CDC captured the fewest males and females. GT captured significantly ($F_{3, 27} = 9.66$; $P = 0.0103$) more gravid females than MM-PRO, MM-X, or CDC.

Mean percentage of variation in sex and physiological state responses among trap types showed the majority of males (58%), females (64%) and blooded females (71%) were captured by MM-PRO, and the majority of blooded-gravid (79%) and gravid females (78%) were captured by GT. CDC captured the lowest percentage of males (7%), females (9%): GT captured the lowest percentage of blooded females (6%), and MM-X captured the lowest percentage (5%) of blooded-gravid and gravid females.

Within trap type, the MM-X collected proportionately more males (22%) than CDC, MM-PRO, or GT (10–11%), whereas CDC, MM-X, and MM-PRO captured proportionately more blooded females (4–6%) than GT (1%). Thirty-eight and 3% of females captured in GT were gravid and blooded-gravid, respectively.

Cx. nigripalpus was the second most frequently trapped species at Hidden Hammock (Table 1). The low mean number of male captures, which did not vary significantly by trap type, ranged from 0 (MM-X) to 0.5 per trap day (GT). Mean capture rates for females did not differ significantly by trap type except for GT, which was significantly lower ($F_{3, 27} = 15.11$; $P < 0.0001$) than MM-Pro, MM-X, and CDC. In general, the average number captured by MM-Pro was 2 times that captured by CDC and 3 times that captured by MM-X. None of the traps captured blooded-gravid female *Cx. nigripalpus*, nor were differences in the numbers of blooded or gravid females collected by trap type significant (although CDC collected 2 or more times as many gravid females as GT, MM-X, or MM-Pro).

Mean percentage of variation in sex and physiological state responses among trap types showed the highest percentage of female *Cx. nigripalpus* was collected by MM-Pro (53%) followed by CDC (28%). GT collected only 1%. Despite low average numbers of blooded females in all traps, 88% of such individuals were collected by counterflow geometry design devices (MM-Pro and MM-X). Fifty-one percent of all the gravid *Cx. nigripalpus* were collected by CDC.

Within trap type, physiological state responses were similar for MM-Pro, MM-X, and CDC. Slightly more than half (58%) of GT collections were made up of male and gravid female *Cx. nigripalpus.*

None of the traps at Hidden Hammock collected male *Cx. erraticus* or *Cx. salinarius*, and none collected ≥1 female of either species, on average, per trap day. The CDC trap collected the most *Cx. erraticus* (0.69 females per day) and the MM-Pro collected the most *Cx. salainarius* (0.13 females per day). There were no specimens of either species that contained blood or eggs in the abdomen.

Swine Unit. *Cx. nigripalpus* was the most frequently trapped species (Table 2). On average, significantly more male *Cx. nigripalpus* ($F_{3, 27} = 8.25$; $P = 0.0006$) were captured by CDC than other trap types. CDC and MM-X captured significantly more females ($F_{3, 27} = 151.60$; $P < 0.0001$) and blooded females ($F_{3, 27} = 19.86$; $P < 0.0001$) than PRO, which captured significantly more females and blooded females than GT.

Table 2. Comparisons of four different trap types for the collection of *Culex* mosquitoes at University of Florida Swine Research Unit, Alachua Co., FL

Species	Sex/physiological state	Mean no. (SE) of mosquitoes collected/trap night for each trap type			
		GT	CDC	MM-X	MM-Pro
Cx. quinquefasciatus	♂/total	9.86 (2.06)a	1.61 (0.34)b	6.96 (2.06)a	6.79 (1.64)a
	♀/total	60.07 (11.79)a	18.64 (4.82)b	84.36 (20.51)a	75.50 (17.82)a
	♀/blooded	1.29 (0.82)a	0.00 (0.00)a	0.04 (0.04)a	2.68 (2.64)a
	♀/blood/gravid	0.64 (0.37)a	0.00 (0.00)a	0.00 (0.00)a	0.00 (0.00)a
	♀/gravid	20.89 (4.38)a	0.00 (0.00)b	0.07 (0.07)b	0.14 (0.07)b
Cx. nigripaplpus	♂/total	1.25 (0.33)a	6.57 (1.59)b	3.36 (0.72)c	3.29 (0.71)c
	♀/total	8.46 (2.47)a	2158.57 (427.38)b	1289.86 (248.29)b	764.86 (169.96)c
	♀/blooded	0.07 (0.05)a	6.39 (1.79)b	6.68 (2.59)b	2.36 (0.94)c
	♀/blood/gravid	0.04 (0.04)a	0.32 (0.19)a	0.68 (0.28)a	0.36 (0.19)a
	♀/gravid	0.93 (0.31)a	1.86 (0.89)a	2.50 (1.19)a	0.46 (0.17)a
Cx. erraticus	♂/total	0.00 (0.00)a	0.04 (0.04)a	0.21 (0.18)a	0.00 (0.00)a
	♀/total	0.11 (0.06)a	25.18 (5.31)b	38.82 (7.01)c	3.79 (0.92)d
	♀/blooded	0.00 (0.00)a	0.04 (0.04)a	0.11 (0.11)a	0.00 (0.00)a
Cx. salinarius	♂/total	0.00 (0.00)a	0.11 (0.06)a	0.11 (0.06)a	0.00 (0.00)a
	♀/total	0.00 (0.00)a	24.25 (5.16)b	27.89 (7.29)b	12.07 (3.06)c

PROC GLM and multiple comparison (Tukey's HSD) test (SAS Institute 1988) performed after $\log_{10}(x + 1)$ transformations. Means within each row (for each sex and physiological state) having the same letter are not significantly different (n = 28 nights, $P \leq 0.05$).

Mean numbers of blooded-gravid and gravid females did not differ significantly among trap types, although mean capture rates for gravid females were highest for MM-X and CDC.

Mean percentage of variation in sex and physiological state responses among trap types showed the majority of male (46%) and female (51%) *Cx. nigripalpus* were captured by CDC. MM-X captured a slightly higher percentage of blooded females (43%) than CDC (41%), and it captured the highest percentage of blooded-gravid (49%) and gravid (43%) females. GT captured the lowest percentage of *Cx. nigripalpus* in all categories, except gravid females (16%), which were captured at twice the rate as MM-Pro.

Within trap type, mean percentage of variation in sex and physiological state responses indicated the GT collected proportionately more male (13%) and proportionately fewer female *Cx. nigripalpus* (87%) than other trap types. The percentage of the total catch comprising blooded, blooded-gravid, and gravid females was >1% for all trap types except for gravid females in GT (11%).

Cx. quinquefasciatus was the second most frequently trapped species at the Swine Unit (Table 2). Significantly fewer males ($F_{3, 27}$ = 9.25; P = 0.0003) and females ($F_{3, 27}$ = 12.21; P < 0.0001) were captured by CDC than the other trap types. The descending order of mean capture rate by trap type was different for males (GT > MM-X > MM-Pro > CDC) than for females (MM-X > MM-Pro > GT > CDC), however. Mean numbers of blooded and blooded-gravid females did not differ significantly by trap type, whereas GT collected significantly more gravid females ($F_{3, 27}$ = 142.83; P < 0.0001) than MM-X, Pro, or CDC.

Among trap types, the lowest percentage of male *Cx. quinquefasciatus* was collected by CDC (6%) and the highest percentage was collected by GT (39%). CDC collected 8% of all females, GT 25% (32% of all blooded females), MM-X 35%, and MM-Pro 32% (including 67% of all blooded females). All blooded-gravid females and >99% of the gravid females were collected by GT.

Within trap type, the GT captured proportionately more male (14%) and proportionately fewer female *Cx. quinquefasciatus* (86%) than other trap types. MM-Pro and GT collections made up 4 and 2% blooded females, respectively, whereas <1% of the females collected by MM-X and CDC were blooded. Blooded-gravid females were captured only by GT, and 35% of the females captured using this trap were gravid (compared with <1% for MM-X, MM-Pro, and CDC).

Cx. erraticus and *Cx. salinarius* also were trapped at the Swine Unit (Table 2). For both species, there was no significant difference in the numbers of males collected by trap type. For *Cx. erraticus* significantly more females ($F_{3, 27}$ = 48.48; P < 0.0001) were captured by MM-X than CDC, by CDC than MM-Pro, and by MM-Pro than GT. Most (>99%) *Cx. erraticus* collected were empty females, regardless of trap type. Only CDC and MM-X captured blooded females (29 and 71%, respectively, of all females in this category), although means for the two trap types were not significantly different. Gravid *Cx. erraticus* were captured only by MM-Pro (0.04 gravid females per trap night). For female *Cx. salinarius*, the descending order of mean capture rate by trap type was MM-X > CDC > MM-Pro (GT did not capture any *Cx. salinarius*). Differences between CDC and MM-X were not significant, but both trap types collected significantly more females ($F_{3, 27}$ = 87.00; P < 0.0001) than MM-Pro. Among trap types, 86 and 14% of all blooded females were only captured by MM-Pro (0.57 females per trap night) and MM-X (0.11 females per trap night); blooded-gravid females were not collected by any trap type, and only MM-Pro captured any gravid females (0.07 females per trap night). Within MM-X, MM-Pro, and CDC trap types, >99% of the catch composition was empty females.

Discussion

These data indicate that the concept that "one size fits all" does not apply to trap efficacy and *Culex* mosquito species. Indeed, trap efficacy differed according to trap type, mosquito species, gonotrophic condition of the female mosquitoes, and the geographic location that the species was collected.

In this study, the GT was a good trap for collecting *Cx quinquefasciatus,* but it was poor or inefficient for collecting *Cx. nigripalpus.* No specimens of *Cx. salinarius* were collected in the GT at either site. It is impossible to tell from this study how efficient the GT is for collecting *Cx. erraticus,* because very few individuals of this species were collected at either site. These data emphasize the caution that should be taken in relying too much on a single surveillance trap technology to make conclusions. Based on GT collections only, the swine unit would be considered as a site dominated by *Cx. quinqufasciatus.* However, based on years of collecting and identifying larvae at both our study sites, Becnel et al. (2001) demonstrated that *Cx. nigripalpus* was the dominant species at the swine unit, especially during the summer. The other three trap types tested clearly verify that *Cx. nigripalpus* is the dominant species at this site. At the dairy site, *Cx. quinquefasciatus* was the dominant species year-round. All four trap types are in agreement with that assessment. Why the GT is inefficient for collecting *Cx. nigripalpus* is unknown. What is known is that the GT works on a totally different concept than the other three trap types, i.e., it was originally developed by Reiter (1983) to sample the gravid female portion of populations of *Cx. pipiens,* which is a member of the same species complex as *Cx. quinquefasciatus.* Because it was developed to sample gravid females it uses an oviposition attractant (hay infusion) not CO_2 to draw the females to the trap. The other three trap types tested use CO_2 as the primary attractant. The host-seeking portion of the population is attracted to CO_2 and thus dominates the collections in these trap types. At both sites, the GT was efficient at collecting *Cx. quinquefasciatus,* especially the gravid and blood/gravid portions of the population. Thus, the GT works well for the species complex, especially the gravid females, that it was originally designed to capture. What changes can be made to make it more attractive to the other *Culex* spp. are currently under investigation. Some aspects being investigated include trap shape, type of infusion, and color and size of pan that holds the infusion.

In contrast to the GT, the CDC trap was overall the most efficient trap for collecting *Cx. nigripalpus* and the least efficient for collecting *Cx. quinquefasciatus.* What makes it more attractive for collecting *Cx. nigripalpus* is a matter of speculation. Trap height above ground level may be one important variable and will be investigated in future studies with *Cx. nigripalpus.* Casual observations (D.L.K.) suggest that *Cx. nigripalpus* is attracted by light, and *Cx. quinquefasciatus* is repelled by light. In a study comparing the MM-X and CDC trap (with or without a light) in Tanzania (Mboera et al. 2000), the MM-X collected significantly more *Cx. quinquefasciatus* than did the CDC with or without the light. Significantly more *Cx. quinquefasciatus* were collected by the CDC trap without light than with light. CDC light traps with or without dry ice have been used extensively to collect *Cx. nigripalpus* in Florida (Boike 1963, Dow 1971, Provost 1969). CDC traps without any attractant consistently caught the least number of mosquitoes. Traps baited with light only did very well, but traps baited with both light and CO_2 did the best.

Overall, for all species combined, the two traps designed with counterflow technology were the most efficient traps. At the dairy site, the MM-Pro collected more of each species except *Cx. erraticus;* but it was the second most efficient trap (the CDC was best) for this species at this site. At the swine unit, the MM-X trap was the most efficient trap for all the *Culex* spp., except *nigripalpus;* it was the second best trap (CDC was the best) at this site for this species. The switch in trap efficiencies between sites is perplexing because many adult *Cx. nigripalpus* and *Cx. quinquefasciatus* were obviously present at both sites.

One possible explanation is some kind of trap–environment interaction (i.e., trap placement, competing visual, chemical, and/or physical attractants and temporal changes [seasonal effects]) may be occurring. Becnel et al. (2001) have documented environmental changes at our two study sites. At the swine unit, these changes seem to have a huge impact on the presence of *Cx. nigripalpus* and *Cx. quinquefasciatus* immature stages abundance. These authors found epizootics of a baculovirus (family *Baculoviridae,* genus *Nulclepolyhedrovirus,* CuniNPV) in all instars of *Culex* spp. larvae. *Cx. quinquefasciatus* was especially sensitive to the presence of this virus, and epizootics resulted in high mortality in this species. The highest infection levels were found during the spring and fall with the lowest infection levels found during the winter. This may explain the absence of *Cx. quinquefasciatus* larvae at this site during the warmer months when the epizootics also occurred. At the dairy site, Becnel et al. (2001) found high larval populations of *Cx. quinquefasciatus* (average of 19,000 $\pm$ 5,000 per sampling day), but larvae infected with CuniNPV were collected on only five sampling days and never at epizootic levels (0.08 $\pm$ 0.06% infection). There was an 80-fold increase in infection of *Cx. quinquefasciatus* larvae when exposed to CuniNPV in swine but not dairy wastewater. These results indicated that there were factors present in the swine wastewater that mediated transmission of the virus. The main factor turned out to be the balance between Mg^{2+} and Ca^{2+}. It was discovered (Becnel et al. 2001) that epizootics of CuniNPV occurred in the swine wastewater site where Mg^{2+}/Ca^{2+} ratios (1.9:0.8) mediated transmission and did not occur in the dairy wastewater site where Mg^{2+}:Ca^{2+} (3.7:3.0) ratios were unfavorable. During the epizootics, buckets which contained "uninfected" hay infusion were placed around the perimeter of the pond at the swine unit. Many egg rafts of both *Cx. nigripalpus* and *Cx. quinquefasciatus* were recovered from these buckets, which developed into healthy adults of both species (Genie White, personal communication). This would indicate that the *Cx. quinquefasciatus* being captured at the swine unit were probably migrating in from surrounding areas, and those captured at the dairy pond were being locally produced. Conversely, *Cx. nigripalpus* larvae were unable to thrive in water with high calcium content, which occurred at the dairy site; therefore, the *Cx. nigripalpus* collected at the dairy site were probably also dispersing into the trap site from surrounding areas. These findings probably would have an impact on adult collections at each site.

There is also some evidence for a trap–environment effect over geographic space in the literature for *Cx. quinquefasciatus.* Meyer (1991) conducted a study along two parallel 6.4-km-long urban (high housing density) to rural (low housing density) transects in Kern County, CA, in which he compared the operational efficiency of a CO_2-baited CDC trap (no model type given but baited with dry ice and operated without light) with the same model of GT used in our study, and similarly baited with hay infusion, for sampling *Cx. quinquefasciatus.* Meyer (1991) found that female *Cx. quinquefasciatus* were effectively sampled by GT in urban subdivisions and by CO_2-baited CDC traps in rural mixed agricultural areas. Our data indicate a similar trend; the GT caught ≈3 times as many female *Cx. quinquefasciatus* females at the more urban site (swine unit site had been encroached upon by student housing apartment complexes) as the CO_2-baited CDC trap. At the rural dairy site, the CDC caught ≈2 times as many females as the GT. Meyer (1991) concluded that this apparent discontinuity in trap efficiency suggests a sampling confoundment introduced by two key environmental factors that affect ovipositional and host-seeking behavior: 1) access to and number of suitable breeding sources, and 2) distribution patterns of avian hosts in urban versus rural zones. This conclusion was partially based on data obtained during a previous study (Reisen et al. 1990) on dispersal and breeding habits of *Cx. quinquefasciatus* that was conducted in the Los Angeles basin, which revealed that within urban subdivisions host-seeking females are evenly dispersed, whereas gravid individuals tend to become aggregated. Neighborhood surveys of individual premises revealed that diurnal resting shelters such as landscape vegetation and vertebrate hosts, primarily resident birds and domestic pets, were more uniformly distributed in comparison with typical rural environs. However, suitable breeding sources were uncommon and extremely localized at 10% of the premises surveyed. The relative uniformity in distribution of competing avian hosts coupled with aggregated oviposition sites apparently reduced attraction to CO_2 traps but increased attraction to gravid traps. This may help explain some site differences between the GT and our CO_2-baited CDC, traps, but it does not explain why the MM-Pro performs better at our rural dairy site than the MM-X, whereas the opposite is true for the more urban swine unit site, because both of these trap types use CO_2 as their primary attractant.

Other possible explanations may be that there are biological differences (plant and animal) between the two study sites, e.g., host types (cows at the dairy site and pigs/hogs at the swine unit) and availability (bird species may differ between the two sites), mosquito dispersal characteristics, population age structure (oviparity), and seasonal effects. Unfortunately, we did not do any bloodmeal analysis to determine host preference during this study. Larger numbers of blooded, blooded-gravid, and gravid *Cx. quinquefasciatus* were collected at the dairy site than at the swine unit. This may indicate a relatively more "stay-at-home" *Cx. quinquefasciatus* population at the dairy site compared with a transient one at the swine unit, as indicated above. Other comparisons of the subpopulations between sites also may provide some useful information. At the dairy site, for example, there were ≈8.5 times more gravid female *Cx. quinquefasciatus* in the GT as in the MM-Pro but 150 times more in the GT at the swine unit compared with the MM-Pro. Perhaps there are some genetic differences between these geographically separated populations that translate into behavioral differences, but this does not seem likely for *Cx. quinquefasciatus*, based on a recent study of this species by Nayar et al. (2003) in Florida. Basically, their study found that genetic variability values between geographic samples from the Florida panhandle and south Florida were not significant. Indeed, they found that the genetic characteristics of the Florida populations of *Cx. quinquefasciatus* are very similar to populations from areas in the United States where ecological conditions are very different. The same cannot be said for *Cx. nigripalpus.* There is an active research project underway at the Florida Medical Entomology Research Laboratory, Vero Beach, FL, that indicates that there may be several genetically and possibly morphologically distinct subspecies of *Cx. nigripalpus* distributed in various parts of Florida.

Too few *Cx. erraticus* or *Cx. salinarius* were captured during this study by any of the trap designs tested to make any meaningful conclusions about relative trap efficiencies. These species tend to be more abundant during the cooler months in North Central Florida. Future studies are planned that will include the time of the year and sites where these species are most abundant.

It is difficult to compare our results with other studies, because only a few published reports exist with any data for any *Culex* species with the newer counterflow trap technologies and these often used octenol, which is often repellent to some species of *Culex* (Kline 1994), as an additional attractant. Burkett et al. (2001) conducted field studies, with emphasis on anopheline vectors of malaria, in the Republic of Korea with two different CDC-type traps (JH model 1012 and ABC-Pro model), both with the standard CM-47 incandescent bulb: two MM-X traps (with and without octenol) and an MM-Pro (with octenol), but no GT. They also used a Shannon trap, NJ light trap, and EPAR Mosquito Killer trap (model MKS-H, Environmental Products and Research, Blytheville, AR). The ABC-Pro model CDC-type trap was operated with the light set to flicker, but the model 1012 was operated with a steady light. Vaidyanathan and Edman (1997) compared 11 trapping methods and found no significant differences in trap collections between the JH and ABC models. They also observed no significant differences in trap collection numbers for ABC traps with bulbs set to steady or flicker. The two ABC traps were baited differently for CO_2. One trap was baited with 1.5 kg of dry ice, and the other trap was baited with CO_2 dispensed from a 9-kg compressed gas cylinder with a controlled release rate of 500 ml/min. Unfortunately, no CO_2 was used with the JH model CDC-type trap. The ABC model CDC-type traps baited with CO_2 (compressed gas and dry ice) and the MM-X trap with no octenol captured significantly more *Cx. pipiens* than did the other traps. Interestingly, those traps baited with octenol as one of the attractants captured significantly fewer *Cx. pipiens.* Likewise no *Cx. orientalis* Edwards were collected in any of the octenol-baited traps. *Culex tritaeniorhynchus* Giles, *Culex bitaeniorhynchus* Giles, and *Culex vegans* Wiedemann were collected in insignificant numbers for analysis. The Shannon trap did not effectively attract *Cx. pipiens.*

In a study conducted in Australia, Johansen et al. (2003) compared the numbers of mosquitoes collected with the standard Encephalitis Vector Surveillance (EVS) (model E67 PR 101, Australian Entomological Supplies, Pty., Ltd., Coorabel, New South Wales, Australia), CDC-type (unspecified model) light trap, MM-X, and Pro traps. The Pro produced CO_2 during combustion of propane, whereas the MM-X, CDC, and EVS traps were baited with a 1-kg block of dry ice. All four traps were supplemented with octenol (at a release rate of $\approx$4.5 mg/h). In addition to CO_2 and octenol, the CDC and EVS traps had 6.3-V, 150-mA and 12-V, 50-mA incandescent bulbs, respectively, as attractants. The larger light bulbs used in the CDC traps were blacked out with permanent marker ink to reduce light, minimizing the collection of moths. In their first trial, the Pro outperformed the CDC, MM-X and EVS traps. In this trial the Pro performed significantly better than the EVS with collections of *Culex* spp. and *Cx. sitiens* subgroup mosquitoes but not significantly better than the CDC and MM-X traps. In a second trial, the Pro and CDC traps collected significantly more *Culex* spp. than the EVS trap but not the MM-X trap. The CDC trap alone was significantly better than the EVS trap at collecting *Cx. sitiens* subgroup mosquitoes. These investigators concluded that the Pro is an attractive trap for virus surveillance in Australia. Because the Pro can function for up to 20 d before requiring a propane refill, it has particular application for remote areas such as the Torres Strait islands.

Collectively, the data obtained from our study and these other cited studies demonstrate the need to conduct field studies in different geographic areas on both trap design and attractant combinations. Although Burkett et al. (2001) found octenol to significantly reduce trap collections of *Cx. pipiens*, other investigators (Becker et al. 1995, Mboera et al. 2000) found no significant difference in *Cx. pipiens* in octenol- versus nonoctenol-baited traps. Such findings provide us the motivation to continue testing different trap types and various configurations of the same trap type for their efficacy in collecting *Culex* mosquitoes. Plans have been developed to investigate various configurations of the MM-Pro trap. For example it would be interesting to see what effect the addition of light, octenol and a combination of light and octenol to a MM-Pro trap will do to *Culex* species composition and abundance compared with the basic trap. These configurations will be tested continuously for at least 1 yr to determine whether there are any seasonal effects on trap efficiency. Research to further improve the effectiveness of trapping systems through the use of avian and equine mosquito attractants also will be conducted.

The introduction and subsequent rapid spread of WNV underscores the vulnerability of the United States to introduced vector-borne pathogens that affect both livestock and humans and the importance for us to develop improved surveillance methods for adult *Culex* mosquitoes. Improved methods for trapping and controlling the mosquito vectors of WNV also will be crucial for detecting and monitoring any future exotic introductions such as Japanese encephalitis and Rift Valley fever.

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